REMARKS

A. Status of the Claims

The Action acknowledges Applicants' election, without traverse, of the Group I invention, which includes claims 1, 3, and 4 as drawn to a prediction of the existence of tamoxifen-resistant breast cancer and drawn to a method for detecting/diagnosing tamoxifen-resistant breast cancer cells/cancer comprising assaying an obtained sample with antibody binding to TIE-2. In this regard, the Action acknowledges the cancellation of claims 6-21, and the withdrawal from consideration of claim 1 as it is drawn to limitations other than those drawn to TIE-2 as well as the withdrawal of claims 2 and 5. Therefore, it is noted that claims 1, 3, and 4, drawn to TIE-2, are currently under prosecution.

B. Specification

The Action indicates that the specification should be amended to reflect the status of the provisional parent application. Applicants draw the examiner's attention to the Preliminary Amendment submitted by Applicants on June 8, 2001, which included an amendment to the specification to recite priority data. The specification has been further amended herein to include language recommended by the examiner regarding the claiming of priority to a provisional application.

C. Declaration

The Action indicates that the declaration is defective because non-dated alterations to the address of Inventor Friedrichs have been made to the declaration, and a new declaration in compliance with 37 C.F.R. §1.67(a) identifying the application by its serial number and filing

date is required. In accordance with 37 C.F.R. §1.67(a)(2), Applicants herein submit a supplemental declaration with corrected address information of Inventor Friedrichs which identifies the entire inventive entity, but is signed only by Inventor Friedrichs since the error relates only to this inventor.

D. Drawings

The brief description of the drawings is objected to because FIG. 1 consists of FIGS. 1A, 1B, and 1C, but neither the drawings nor the brief description describes FIGS. 1A, 1B, or 1C. Applicants have herein amended the specification to recite a description of FIGS. 1A, 1B, and 1C depicted in the drawings. In particular, the brief description of the drawings in the specification has been amended to indicate that FIGS. 1A, 1B, and 1C show the three bivariate log-log scatterplots that arise from pairwise comparisons of expression data for 588 genes, collected from estrogen-stimulated (ES), tamoxifen-sensitive (TS) and tamoxifen-resistant (TR) breast cancers. FIG. 1A is ES v. TS; FIG. 1B is ES v. TR; FIG. 1C is TS v. TR.

E. The Enablement Rejections Under 35 U.S.C. §112, First Paragraph, are Overcome

1. Nature of the Rejection

Claims 1, 3, and 4 are rejected under 35 U.S.C. §112, first paragraph, because the Action indicates that the specification does not reasonably provide enablement for a method of detecting tamoxifen-resistant breast cancer cells comprising assaying for TIE-2 polypeptide. In particular, the Action indicates that "given the unexpected nature of the results, given that the specification clearly states that this is the first time that a correlation between expression levels for angiogenic

factors and receptors and tamoxifen-resistant breast cancer has been reported, given the well known differences between cultured cell line derived tumor cells and primary tumor cells, given the known artifactural [sic] nature of cell lines, given that the detected polypeptide does not appear to be TIE-2, but rather a putative TIE-2 related protein observed in a cell-line derived tumor, given the art recognized necessity to validate cancer markers in order to determine if they in fact do what is suggested, it cannot be predicted and one of ordinary skill in the art would not believe that it is more likely than not that the invention will function as claimed based only on the MCF-7 tumor model presented." Office Action, page 10, paragraph 1 through page 11, paragraph 1. Therefore, the specification is said to provide "insufficient guidance" with regard to these issues and "provides no working examples which would provide guidance to one skilled in the art" such that one of ordinary skill in the art would be able to practice the claimed invention with a reasonable expectation of success without undue experimentation. Office Action, page 11, paragraph 1. Applicants traverse this rejection.

2. *Enablement Law*

The test of enablement is whether the disclosure, when filed, contains sufficient information regarding the subject matter of the claims as to enable one skilled in the pertinent art to make and use the claimed invention without undue experimentation. *Manual of Patent Examining Procedure (MPEP)* §2164.01, citing *In re Wands,* 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Circ. 1988). In considering whether there is sufficient evidence to meet the enablement requirement has been met, various factors are considered, including (a) the breadth of the claims; (b) the nature of the invention; (c) the state of the prior art; (d) the level of one of ordinary skill; (e) the level of predictability in the art; (f) the amount of direction provided by the inventor; (g) the existence of working examples; and (h) the quantity of experimentation need to make or use the invention based on the content of the disclosure. *MPEP* §2164.01(a) citing *In re Wandts*, 858 F.2d 737, 8 USPQ2d 1404. The examiner's analysis must consider all of the evidence related to each of these factors, and any conclusion of nonenablement must be based on the evidence as a whole. *Id.*

3. *The Specification Contains Information Regarding the Subject Matter of the Claims that is Sufficient to Support Enablement*

The specification as written contains substantial information regarding the subject matter of the claims that is more than sufficient to enable one skilled in the art to make and use the claimed invention without undue experimentation.

The claims are drawn to methods of detecting tamoxifen-resistant breast cancer cells that involve assaying for TIE-2 polypeptide. This includes methods of detecting tamoxifen-resistant breast cancer cell from any species, including humans, by detecting any TIE-2 related polypeptide.

Applicants' specification discloses substantial information regarding tamoxifen resistance on page 2, lines 14 - page 3, line 13. One of ordinary skill in the art would also be very familiar with additional information pertaining to tamoxifen resistance. Applicants' specification notes that the differential expression of certain genes, including TIE-2, are associated with tamoxifen-resistant breast cancer. Specification, page 3, lines 16-26. The results regarding TIE-2 provide the basis for methods directed toward the expression levels of TIE-2 in breast tissue samples which will have utility for the diagnosis and prediction of tamoxifen-resistant breast cancer. Specification, page 3, lines 28-31.

Regarding the particular methodologies to be used to detecting expression levels of TIE-2, immunological methods may be used, including the use of Western blots, immunohistochemistry, ELISA, and other well known techniques for antibody assay of protein expression. Specification, page 4, lines 1-5. Other methods involve use of such antibodies for breast cancer detection, diagnosis, and prediction, by comparing levels of TIE-2 polypeptide in suspected tamoxifen-resistant cancer cells with levels present in groups of known estrogen stimulated, tamoxifen-sensitive, and tamoxifen-resistant breast cancer cells. Specification, page 4, lines 6-10. FIG. 7 demonstrate results of a Western blot analysis using TIE-2 antibody, showing that only tamoxifen-resistant tumors exhibited detectable expression of a high molecular weight (220 kDa) form of TIE-2 (putative TIE-2 related protein).

A discussion pertaining to the definition of TIE-2 protein and TIE-2 gene is provided on page 8, line 11 through page 10, line 4 of the specification. Further, the GenBank Accession No. of the gene encoding human TIE-2 is provided in the specification on page 3, line 19.

Whether a protein is a TIE-2 protein is defined in part by whether the protein in question has the ability to inhibit angiogenesis, or to prevent metastasis or invasive tumor growth.

Specification, page 8, lines 2-4. Molecules possessing this activity may be identified using assays familiar to those of skill in the art. Specification, page 8, lines 5-6. The present invention also relates to fragments of the polypeptides that may or may not retain the angiogenic (or other) activity of TIE-2. See specification, page 9, lines 15-17. Thus, as set forth in the specification, the definition of TIE-2 protein encompasses putative TIE-2 related proteins of a high molecular weight, such as 220 kDa.

Because the methods claimed herein may involve purification of TIE-2, methods pertaining to protein purification are addressed in detail on page 10, line 7 through page 13, line 6 of the specification. One of ordinary skill in the art would be familiar with these and other techniques that can be used to purify proteins such as TIE-2. Information about the use of TIE-2 proteins or peptides as antigens for the immunization of animals for the production of antibodies is provided on page 13, line 25 through page 14, line 5. Techniques pertaining to the generation of antibodies reactive with TIE-2 are provided on page 38, line 28 through page 43, line 27 of the specification.

The specification also includes substantial information pertaining to methods for detecting variation in the expression of TIE-2. See, *e.g.*, Specification, page 44, line 10 through page 40, line 12. Gene amplification and detection methods are also discussed in detail in the specification. Specification, page 46, line 1 through page 56, line 6. Further, techniques pertaining to *in vitro* and *in vivo* assays are included on page 56, line 8 through page 58, line 10.

The Examples demonstrate preferred embodiments of the invention. Disclosed are materials and methods wherein MCF-7-derived tumors are assayed to demonstrate expression of TIE-2. Specification, page 70, lines 1-24. The tumors were assayed by Western blot analysis. Specification, page 70, line 26 through page 71, line 10. The results demonstrate that expression

of TIE-2 mRNA and protein are upregulated in tamoxifen resistant MCF-7 cell line tumor as compared to tamoxifen-sensitive and estrogen stimulated MCF-7 cell line cancers. Specification, page 76, line 6 through page 77, line 31. These results demonstrate that TIE-2 is a positive marker for tamoxifen-resistant breast cancer, and assays for increased expression of this marker may be used to differentiate between tamoxifen-resistant and tamoxifen-sensitive forms of breast cancer. Specification, page 77, lines 8-15.

The entire specification, and in particular the information cited above, is sufficient to enable one skilled in the art to make and use the claimed invention without undue experimentation.

4. The Specification Supports Enablement of Claims Directed to In Vivo Methods of Detecting Tamoxifen-Resistant Breast Cancer Cells

The Action appears to assert that the specification fails to support enablement of the claims to the extent that the claims pertain to *in vivo* conditions. More particularly, the Action notes that "[o]ne cannot extrapolate the teaching of the specification to the scope of the claims because the invention is based on tumor cell line data that is not commensurate in scope with the *in vivo* conditions." Office Action, page 6, paragraph 2. In support of this assertion, certain references are cited to demonstrate that cultured cells exhibit characteristics difference from those cell characteristics noted *in vivo*.

Applicants first note that the examples included in the specification to demonstrate preferred embodiments are examples pertaining to *in vivo* conditions involving mice, and not *in vitro* conditions. The tumor tissue used in the experimental protocol was grown in mice that had been innoculated with MCP-7 tumor cells, and not grown in a petri dish. In particular, the

studies involved tamoxifen-resistant MCF-7 tumors grown in the mammary fat pads of athymic nude mice supplemented with an estrogen pellet. Specification, page 70, lines 2-11. Following removal of estrogen pellets, animals were treated with tamoxifen. After initial growth suppression, the tumors became resistant and growth resumed. Specification, page 70, lines 7-8. Animals were sacrificed at various times to obtain cells for subsequent studies pertaining to TIE-2 expression. Thus, the tumor tissue used in the analyses set forth in the Examples was obtained from mice demonstrating tumor growth, and not from tumor cells that had merely been grown in culture. Thus, the examples disclosed in Applicants' specification pertain to *in vivo* experimental conditions. Therefore, the argument set forth in the Action that the examples do not support claims directed to *in vivo* methods is incorrect.

Perhaps the Action, by citing various references addressing differences between tumor cells grown *in vitro* and *in vivo*, may be attempting to argue that tumors that develop as the result of cell line inoculation into animals have different characteristics than primary animal tumors. If this is the case, then the burden to demonstrate lack of enablement based on this line of argumentation has not been met since the Action does not present any studies demonstrating any such differences.

At most, the Action appears to be arguing that the examples disclosed in Applicants' specification are insufficient to support the claims because the examples do not disclose each and every condition under which tamoxifen-resistant breast cancer develops. However, the existing examples are more than sufficient to support enablement of the claimed invention. Those of skill in the art understand conditions under which tamoxifen-resistant breast cancer develops. Furthermore, the claims at issue pertain to assaying tamoxifen-resistant breast cancer, and not

understanding how tamoxifen-resistant breast cancer develops or treating tamoxifen-resistant breast cancer. Therefore, the standard for enablement should be lower.

According to *MPEP* §2164.02, "[c]ompliance with the enablement requirement of 35 U.S.C. §112, first paragraph, does not turn on whether an example is disclosed." Further, "the specification need not contain an example if the invention is otherwise disclosed in such a manner that one skilled in the art would be able to practice it without an undue amount of experimentation. *MPEP* §2164.02 citing *In re Borkowski,* 422 F.2d 904, 908, 164 USPQ 642, 645 (CCPA 1970). The existing examples, in addition to the remainder of the specification, is more than sufficient to support enablement of the invention as claimed.

5. *There is no Evidence that the Presence of High Molecular Weight Putative TIE-2 Protein is a Cell Line Artifact*

The Action alleges that the difference in the TIE-2 protein found in HuVec cells (140 kDa protein) compared to the 220 kDa high molecular weight form of a putative TIE-2 related protein found in the tamoxifen-resistant breast cancer tissue (Specification, page 78, line 6 through page 79, line 31) was the result of a cell line-related artifact due to change in the chromosomal constitution of the cells used to generate the data, rather than a change associated with tamoxifen-resistant breast cancer. Applicants disagree.

As discussed above, the breast cancer tissue used in the Examples was taken from mice that demonstrated tumor growth following inoculation with MCF-7 cells. As discussed above, the Action has failed to set forth any evidence that tumors grown in animals following inoculation of a cell culture line demonstrate characteristics that differ from the primary tumor.

Furthermore, the results in Example 4 demonstrate that TIE-2 showed increased expression in tamoxifen-resistant tumors, compared to tamoxifen-sensitive and estrogen-stimulated breast cancers. Specification, page 77, lines 3-7. The higher molecular weight form of a putative TIE-2 protein was observed only in the tamoxifen-resistant tumors. As detailed in the specification, all tissue samples were taken from mice that had been innoculated with MCF-7 cells. Specification, page 70, lines 8-11. Had the high molecular weight form of TIE-2 been somehow related to the use of MCF-7 cells in the experimental protocol, then one would have expected the high molecular weight form of TIE-2 to be identified not only in the tamoxifen-resistant tumors, but also in the tamoxifen-sensitive tumors and estrogen-stimulated breast cancers. However, the high molecular weight form of TIE-2 was only identified in tamoxifen-resistant tumors. This finding supports the concept that TIE-2 is a positive marker for tamoxifen-resistant breast cancer, rather than a cell line artifact. Specification, page 77, lines 3-9.

The allegation set forth in the Action that the difference between the 140 kDa TIE-2 protein in HuVec cells compared to the 220 kDa high molecular weight putative TIE-2 related protein found in tamoxifen-resistant breast cancer is the result of a cell line-related artifact is mere speculation on the part of the Examiner. No reasonable basis has been set forth in the Action for believing that this difference is the result of a cell line-related artifact. As set forth above, the tissue used in the animal studies was grown in mice *in vivo*, and not in cell culture. Furthermore, the Action has failed to meet the burden of proof unless proof is set forth demonstrating differences in TIE-2 between tissue grown *in vitro* and tissue grown *in vivo*.

6. *The Action Fails to Demonstrate that Overexpression of the TIE-2 RNA Gene Product Does Not Predict Overexpression of TIE-2 Protein*

The Action argues that "even if the RNA gene product of the TIE-2 gene were to be overexpressed, it could neither be predicted nor would it be expected that a similar overexpression of protein would also be found because evidence abounds in which protein levels do not correlate with steady-state mRNA levels or alterations in mRNA levels in cancer tissues." Office Action, page 8, paragraph 2. In support of this notion, the Action cites (1) Hell *et al.* (Laboratory Investigation, 73:492-496, 1995), a study pertaining to Bcl-2 protein expression in Hodgkin's disease; (2) Fu *et al.* (EMBO Journal, 15:4392-4401, 1996), a study pertaining to p53 protein expression in acute myelogenous leukemia; and (3) Jang *et al.* (Clinical and Experimental Metastasis, 15:469-483, 1997), a study which is said to teach that further studies are necessary to determine if changes in protein levels track with changes in mRNA levels for metastasis associated genes in murine tumor cells.

The above "abounding evidence" fails to support the notion that overexpression of TIE-2 mRNA gene product does not correlate with TIE-2 protein expression. In particular, none of the studies cited in the Action pertain to breast cancer or the TIE-2 protein.

It is well known in the art that increases in mRNA levels are often associated with an parallel increase in expression of the associated protein. The presence of a few exceptions in the vast field of oncology, as cited in the Action, does not support lack of enablement for the claimed invention.

In order to make a rejection based on lack of enablement, the examiner has the initial burden to establish a ***reasonable basis*** to question the enablement provided for the claimed invention. *MPEP* §2164.04, citing *In re Wright*, 999 F.2d 1557, 1562, 27 USPQ2d 1510, 1513 (Fed. Cir. 1993) (emphasis added). The references cited by the examiner, which neither pertain

to breast cancer nor to TIE-2, fail to qualify as a reasonable basis for establishing a basis for lack of enablement.

As noted above, the determination of enablement is based on the evidence as a whole. *MPEP* §2164.05. The evidence provided by the Applicant need not be conclusive, but merely convincing to one skilled in the art. Applicants' specification, including the working examples discussed above, provide evidence that would be sufficient to convince one skilled that they would be able to make and used the claimed invention, which is sufficient to meet the enablement requirement.

7. The Examples in Applicants' Specification are Sufficient to Support the Invention as Claimed

According to the Action, "one cannot extrapolate the teachings of the specification to the scope of the claims because this single example, drawn to a cell culture tumor model has not established that the overexpression of TIE-2 is a 'marker' for tamoxifen-resistant breast cancer." Office Action, page 9, paragraph 2. In support of this notion, the Action cites Tockman *et al.* (Cancer Res., 52:2711s-2718s, 1992), which is said to teach that "prior to successful application of newly described markers, research must validate the markers against acknowledged disease end points, establish quantitative criteria for marker presence/absence and confirm marker predictive value in prospective population trials." Office Action, page 9, paragraph 2.

It should first be noted that Tockman *et al.* is a reference that pertains to biomarkers for early lung cancer detection. It does not address biomarkers for breast cancer.

Whether or not Tockman *et al.* should be applied to biomarkers for cancers other than lung cancer is inapplicable to the issue at hand. The issue is whether Applicants' specification as written contains information regarding the subject matter of the claims that is sufficient to enable

one skilled in the art to make and use the claimed invention without undue experimentation. As discussed above, the Examples set forth in the specification provide experimental data demonstrating that TIE-2 is a positive marker for tamoxifen-resistant breast cancer. These Examples, along with other parts of the specification set forth in detail above, are more than sufficient to teach one skilled in the art how to practice the claimed invention without undue experimentation.

The Action also appears to require that human data be provided in the Examples since use of the claimed invention for determination of tamoxifen-resistant breast cancer in humans is contemplated. However, this is not the case. As noted above, "[t]he specification need not contain an example if the invention is otherwise disclosed in such a manner that one skilled in the art will be able to practice it without undue experimentation." *In re Borkowski*, 442 F.2d 904, 908, 164 USPQ 642, 645 (CCPA 1970). The specification as written, including the data pertaining to the mouse model of breast cancer disclosed in the specification, is sufficient to enable one skilled in the art to practice the invention without undue experimentation.

Furthermore, the *MPEP* notes that "[f]or a claimed genus, representative examples together with a statement applicable to the genus will ordinarily be sufficient if one skilled in the art (in view of the level of skill, state of the art and the information in the specification) would expect the claimed genus could be used in that manner without undue experimentation." *MPEP* §2164.02. "Proof of enablement will be required for other members of the claimed genus only where adequate reasons are advanced by the examiner to establish that a person skilled in the art could not use the genus as a whole without undue experimentation." *Id.*

In the instant case, the claimed genus encompasses methods of detecting tamoxifen-resistant breast cancer cells in humans. Applicants have provided sufficient information

pertaining to representative examples, as discussed above. Further, one of skill in the art would understand, from reading the specification, that the claims encompass methods of detecting tamoxifen-resistant breast cancer cells in humans. See, *e.g.*, Specification, page 1, line 20 through page 2, line 6. In addition, as discussed above, the reasons set forth in the Action are insufficient to demonstrate that one of skill in the art would not be able to practice the claimed invention as it is applied to humans without undue experimentation. Therefore, Applicants do not have the burden of providing examples using human data.

8. The Action Admits that Claims 22-25 are Enabled

According to the Action, "the specification, ***while being enabling for a method of detecting tamoxifen-resistant MCF-7 breast cancer cells comprising assaying for the overexpression of a high molecular weight, 220 kDa putative TIE-2 related polypeptide***, does not reasonably provide enablement for a method of detecting tamoxifen-resistant breast cancer cells comprising assaying for TIE-2 polypeptide." Office Action, page 3, last paragraph through page 4, first paragraph (emphasis added).

Claim 22 in the amendment set forth herein pertains to a method of detecting tamoxifen-resistant breast cancer cells that involves contacting the sample suspected of containing tamoxifen-resistant breast cancer cells with an antibody that specifically binds to a TIE-2 polypeptide with a molecular weight of about kDa. Claim 23, which depends from claim 22, includes the limitation wherein the tamoxifen-resistant breast cancer cells are MCF-7 cells. Claim 24, which also depends from claim 22, includes the limitation of further comprising providing a diagnosis of tamoxifen-sensitive or tamoxifen-resistant breast cancer. Claim 25, which also depends from claim 22, includes the limitation of further comprising providing a

prediction of the existence or development of tamoxifen-resistant breast cancer.

As noted in the Action, these claims are fully enabled by the instant specification. See, *e.g.*, those sections of the Specification cited above in subpart 4 of this section of the Response. By including these claims in the Amendment disclosed herein, Applicants in no way concede that the claims as originally written are not enabled.

9. Conclusion

For the reasons set forth above, the specification contains sufficient information regarding the subject matter of the claims as to enable one skilled in the art to make and use the claimed invention without undue experimentation. Therefore, the enablement rejections under 35 U.S.C. §112, first paragraph, are overcome.

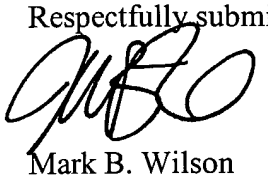
F. Petition for Extension of Time

Pursuant to 37 C.F.R. § 1.136(a), Applicant petition for an extension of time of one month up to and including February 17, 2004 in which to respond to the Office Action dated October 14, 2003. If the check is inadvertently omitted, or the amount insufficient, or should any additional fees under 37 C.F.R. §§ 1.16 to 1.21 required for any reason relating to the enclosed materials, or should an overpayment be included, the Commissioner is authorized to deduct or credit the appropriate fees from or to Fulbright & Jaworski Deposit Account No.: 50-1212/UTSK:348US.

G. Conclusion

In view of the foregoing, it is believed that all claims are in condition for allowance, and a Notice of Allowance is earnestly solicited. The Examiner is invited to contact the undersigned attorney at 512-536-3035 with any questions, comments or suggestions relating to the referenced patent application.

Respectfully submitted,



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